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Developing a Protocol for Creating Microfluidic Devices with a 3D Printer

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Peer Review

This work has undergone a double-blind review by a minimum of two faculty members from institutions of higher learning from around the world. The faculty reviewers have expertise in disciplines closely related to those represented by this work. If possible, the work was also reviewed by undergraduates in collaboration with the faculty reviewers.

Abstract

Microfluidics devices have high importance in fields such as bioanalysis because these devices have the ability to manipulate small volumes of fluid, typically ranging from microliters to picoliters. Small samples of fluids can be quickly and easily tested using reactions performed with complex microfluidic devices. Many methods have been previously developed to create these devices, including traditional nano-lithography techniques borrowed from the field of microelectronics. However, these traditional techniques are cost-prohibitive for many small-scale laboratories. This research explores a relatively low-cost technique using a 3D printed master, which is used as a template for the fabrication of polydimethylsiloxane (PDMS) microfluidic devices. The masters are designed using computer aided design (CAD) software and can be printed and modified relatively quickly. We have developed a protocol for creating simple microfluidic devices using a 3D printer and PDMS adhered to glass. We have also explored methods to overcome the size-limits of the 3D-printed master templates by using shrinkable polymers and modified channel geometries to create a flow-focusing channel. This relatively simple and lower-cost technique can now be scaled to more complicated device designs and applications.

Keywords

microfluidic, 3D printer, rapid prototype

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INTRODUCTION

Microfluidic devices are used in fields such as bioanalysis, materials engineering, and chemistry (Femmer et al. 2015; Gross et al. 2014; Shah et al. 2008; Shirk et al. 2013; Utada et al. 2007; Ye et al. 2015). These devices are especially useful in bioanalysis because they have the ability to manipulate relatively small volumes of fluid, requiring small samples from a test subject. Traditionally, these devices have been made by borrowing techniques from the world of micro- and nanofabrication, such as photolithography (Whitesides and Stroock 2001). Photolithography involves coating a sample substrate in photoresist, a photo sensitive chemical which becomes soluble when exposed to UV radiation. Next a photomask is affixed to the sample, which is then exposed to UV light. Upon development, the exposed resist will be washed away, leaving behind the desired pattern. While reliable, this technique can be cost prohibitive for small-scale production (Plummer et al. 2000). Other researchers, such as Grimes, et al. (2008) have used novel and inexpensive templates made from "Shrinky-Dinks". We instead chose to use a consumer-grade 3D printer.

The popularization and commercial availability of 3D printers has opened up a new avenue for device production. A 3D printer works by interpreting a three dimensional computer drawing of an object and slicing the drawing into layers. The printer then creates the object by extruding heated material and printing the object layer by layer. In our case, the extruded material is acrylonitrile butadiene styrene (ABS plastic). Using a 3D printer, templates for microfluidic device channels can be designed, modified and printed relatively quickly. The templates can then be cast with polydimethylsiloxane (PDMS) and subsequently cured. The PDMS mold can be removed from the master and adhered to a glass slide, creating the fourth wall of the microfluidic channels (Au et al. 2016; Bishop et al. 2015; Kitson et al. 2012; Lee et al. 2014; McDonald et al. 2002; Rogers et al. 2015; Whitesides and Stroock 2001).

Use of a 3D printer reduces cost, making PDMS microfluidic devices more accessible to small-scale production. 3D-printed templates also allow for accelerated testing and modification of device designs; modifications can be added to the template file and printed out immediately (Gross et al. 2014; O'Neill et al. 2014).

MacDonald, et al. (2002) have demonstrated a method to create microfluidic devices in PDMS using solid object printing. We seek to duplicate that work with our own printer and propose a double casting technique to overcome the resolution limitations of the 3D printer.

METHODS

Template Design

Templates for microfluidic devices were designed using Rhinoceros 5.0 computer aided design (CAD) software. A two dimensional outline of the channel structure was created, and then the drawing was extruded in the third dimension to give the channels depth. Test templates were designed with a variety of features, including curved and straight channels. The design file was then exported as a stereolithography (.stl) file-type to the 3D printer.

3D Printing

Templates were printed using a MakerBot Replicator 2X Experimental 3D Printer. This is a commercially available printer with dual extruder capabilities. The printer uses proprietary software, MakerBot

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Desktop, which divides the 3D file into layers and communicates printing instructions to the printer. A 3D object is printed by heating and extruding plastic through a nozzle, which has two degrees of movement, onto a heated platform. The platform descends away from the nozzle after each layer is printed, creating the third dimension. The resolution can be adjusted to create finer layers and increase the detail of the object, but at the cost of speed of printing. The MakerBot Desktop software allows the user to further scale a 3D design, so that a number of templates of various sizes may be created using just one design file.

Additionally, the MakerBot printer and software have a number of preference settings that affect the final product. Specifying the number of shells, or outlines, around a 3D-printed shape affects the quality and resolution of the print, as well as the speed of printing. Rafts can be used to add an additional layer underneath the 3D-printed structure. Supports are removable structures that support an overhanging part as it is printed. Finally, the infill setting affects the volume inside of a part that is filled with solid material, and this feature is used to save weight and material when printing a more massive part. Typically, our templates were printed with a single shell and the default infill setting, and no rafts or supports were needed.

Creating a PDMS and Glass Microfluidic Device – Single Casting Technique

Microfluidic devices were created from Sylgard 184 polydimethylsiloxane (PDMS) [Dow Corning, Auburn, MI] using the printed template as negative mold to create channels. The template is adhered to the bottom of a glass dish and the PDMS solution is poured over the template. Sylgard 184 PDMS is a two-part silicone that has a lower viscosity when first mixed. The silicone was mixed and poured carefully to reduce chances of gas bubbles, and gas bubbles were eliminated in early stages of curing using the tip of a razor blade. It takes approximately 48 hours to cure at standard temperature and pressure. The curing process can be accelerated by placing the silicone into a heated environment. At 40 °C, the curing time is reduced to 2 hours.

Once cured, the PDMS microfluidic device is de-molded from the 3D-printed template, and the excess PDMS is cut away from the sides of the template. The PDMS may be adhered directly to a cleaned piece of glass to add a fourth side to the channel walls created by the template. It is necessary to chemically alter the surface of the PDMS with oxygen plasma to encourage permanent bonding with the glass. This was attempted with a microwave-assisted plasma chamber created in a vacuum container, as well as with a telsa-coil device. In theory, the unsatisfied bonds in both the PDMS and the cleaned glass should create permanent adhesion. In practice, the devices required additional, chemical, adhesive to operate as microfluidic devices. Commercially available adhesives Devcon Home 5 Minute Epoxy Gel [Illinois Tool Works, Riviera Beach, FL], Loctite Extra Time Epoxy [Henkel Consumer Adhesives, Avon, OH], Loctite GO2 Glue [Henkel Corporation, Rocky Hill, CT], and Loctite Epoxy Instant Mix [Henkel Corporation, Rocky Hill, CT] were tested.

Creating a PDMS and Glass Microfluidic Device – Double Casting Technique with Shrinking Silicone

The resolution limit of the MakerBot 3D printer was approximately 0.2 mm. To create devices with size scales smaller than the resolution of the 3D printer, positive templates were printed and a secondary negative template was created with a silicone rubber that shrank as it cured: Mold Star 16 Platinum Silicon Rubber with the addition of Novocs Gloss Silicone Solvent [Smooth-on, Macungie, PA]. The secondary negative template created with this shrinking silicone was de-molded after two hours and allowed to continue curing and shrinking for two days. The shrunken mold can then be used to make sub- sequent molds using double casting techniques. When desired dimensions have been reached, the template could then be cast using the Sylgard 184 PDMS and a releasing agent to create a device as described previously. However, this step has not yet been successfully completed by this research team.

RESULTS AND DISCUSSION

Bonding PDMS to Glass Substrate

A marked improvement in PDMS-glass bonding was noticed when the surface roughness produced by the 3D printed master was minimized. This can be achieved through heat or exposing the ABS plastic template to acetone vapor.

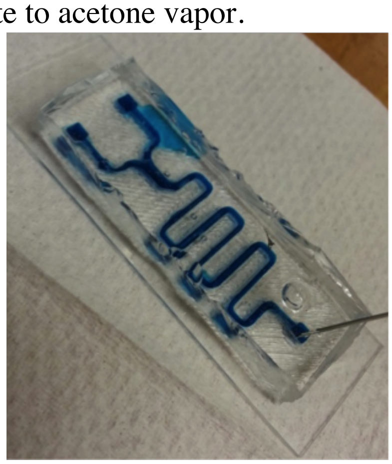

Figure 1. Single-cast PDMS microfluidic device adhered to glass. The small leak of blue-dyed water is due to imperfect adhesion. The glass is a standard three by one inch microscope slide.

However, both of these methods cause blurring and loss of detail of thin lines and fine features that are desirable for narrow channel widths. The best PDMS-glass bonds occurred with the addition of an adhesive.

Single Casting Technique

Using the Makerbot Replicator 2X and the simplest single casting technique, we were able to achieve channel width dimensions under 0.3 mm, similar to dimensions that other researchers, such as McDonald et al. (2002), have reported. An early prototype device is shown in Figure 1. Using photographs from optical microscopy, we measured a difference in channel width between the top of a PDMS channel and the bottom of the channel, visible in the optical micrograph and further illustrated in the schematic in Figure 2. This indicates a defect in the channels because the cross section is not square like the original master templates. This may be due to the Sylgard 184 PDMS shrinking as it cures.

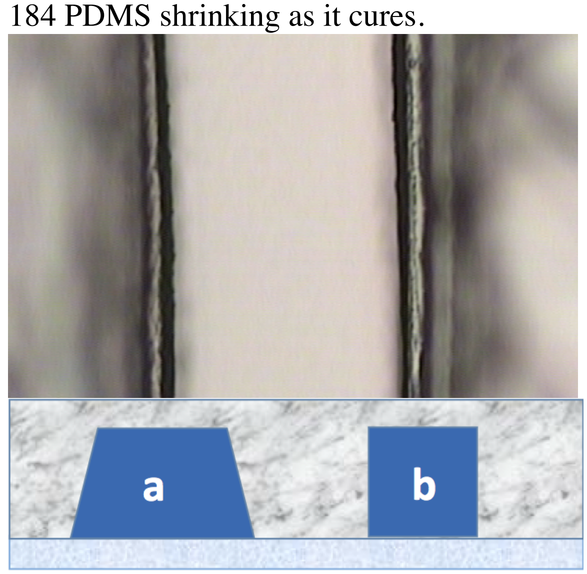

Figure 2. Channel geometry imperfections. Top: Optical micrograph of PDMS microfluidic channel. Channel is approximately 0.3 mm wide. Bottom: Illustrated cross-section of experiment PDMS channels (a) and theoretical template cross-section (b).

Double Casting Technique with Shrinking Silicone

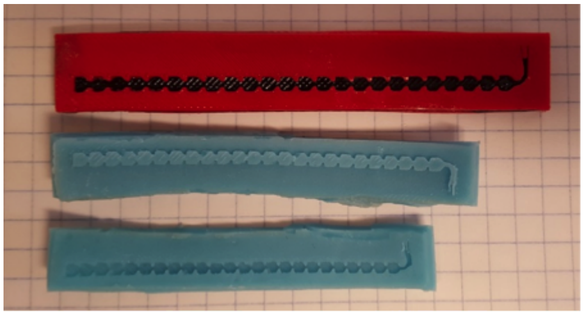

Figure 3. Shrinking Silicone Device Templates. Top: 3D-printed master template; Middle and Bottom: Prototype double-cast silicone device at various stages of shrinking. The graph paper background is a one cm square for scale.

Our experiments using a double casting technique with shrinkable silicones indicated that this is a viable method of building PDMS microfluidic devices with even smaller features than devices fabricated with the single casting technique. We were able to achieve shrinkage of approximately 20% with each casting. However, we saw some anisotropic behavior with the shrinking silicone. It was found that the depth of Mold Star silicone should be at least 10% of the larger length or width dimension to prevent warping near the edges. Figure 3 shows a photograph of the 3D-printed positive template, a shrunken negative silicone mold, and a shrunken positive silicone mold.

The dynamics of how the Mold Star Series Silicone Rubber shrinks pertinent geometries was characterized as well as the time scale for shrinking. A series of lines and shapes were 3D-printed and cast with the Mold Star silicone. These geometries were measured each day as the silicone shrunk. It was determined that the mold shrinks rapidly for the first two days and then shrinks much more slowly for an additional five days as seen in Figure 4. Thus, an optimal shrinking time is 2 days, which results in a mold shrunk by nearly 20%. Longer times give further shrinking,

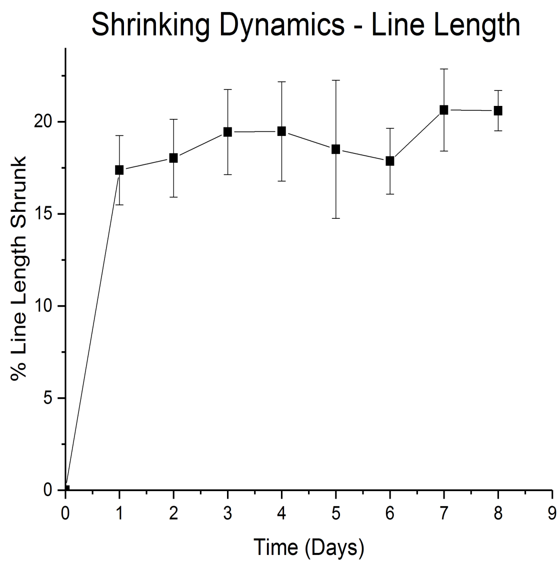

Figure 4. Measurements of shrinking silicone (Mold Star) cured at room temperature and pressure. The length of various test lines were measured over seven days. The percent shrunk each day in comparison with the original length was calculated.

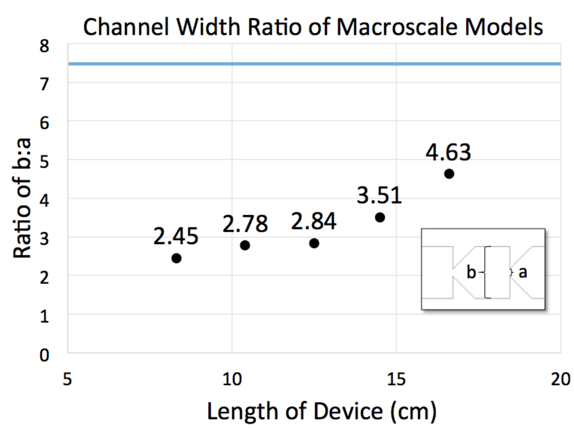

Figure 5. The ratio of the widest channel width at "b" to the narrowest width at "a" is plotted as a function of the device length. The design ratio for this device is 7.5, which is indicated by the solid blue line. The inset schematic illustrates the channel geometries that were measured.

but at the cost of rapid production.

Additionally, five macroscale devices, ranging in sizes from approximately 8 cm to 17 cm in length, were successfully fabricated using the double casting technique. These devices had more complex channel geometries than the device shown in Figure 1. The macroscale devices were designed with a specific ratio between the narrowest and widest portions of the channels, indicated by the solid blue line in Figure 5. As subsequent devices were made with the double casting technique, the ratio of channel width to channel height was reduced. The ratio between the channel widths can be seen in Figure 5 for the five different device sizes; the inset schematic shows the geometries that were measured.

CONCLUSIONS

Based on the results we have compiled, a microfluidic device fabricated in this manner is very feasible. 3D-printed templates allow for quick turn-around in the design, build, test engineering cycle. We have also managed to overcome some size limitations using a double casting technique and shrinking silicones.

At this point, we continue work to optimize the fabrication process, including surface roughness and adhesion issues. While optimization continues, the feasibility of production for multilevel and more complex devices will be explored.

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